Scientific Abstract

In this phase I/II clinical trial, increasing doses of a DNA plasmid called Synchrotope MA2M which encodes two epitope peptides from the melanoma antigen Melan-A/MART-l, amino acids 26-35 and 31-70, will be injected intra-lymphnodally in patients with stage IV metastatic melanoma in order to define the toxicities and MTD (if any) as the primary endpoints and to determine whether there has been an immune response to the DNA vaccine directed against Melan-A/MART-l. Although measurable disease will not be a requirement for study entry, anti-tumor responses will be assessed in patients with measurable disease. The hypothesis of this study is that the Synchrotope MA2M plasmid DNA vaccine is safe and tolerable when used for the treatment of stage IV malignant melanoma and result in the generation of an immune response directed against Melan-A/MART-l-expressing tumor cells.

The population treated with the DNA vaccine will include patients with stage IV metastatic melanoma without brain metastases who are HLA-A2 positive, since the epitopes encoded by the plasmid are restricted to HLA-A2. Expression of Melan-A/MART-l in the tumor will also be required for entry. Increasing doses of the DNA plasmid vaccine starting at 500 μg per dose and increasing to 1000 μg then 1500 μg per dose in cohorts of six patients per dose will be administered by continuous infusion using a miniaturized pump via a catheter inserted into a groin lymph node under ultrasound guidance. The continuous infusion of 96 hours, followed by removal of the catheter and a nine-day rest period, comprises one cycle lasting two weeks. Proper catheter position will be verified by ultrasound on the first and at the end of the fourth day of infusion of each cycle. Four cycles of two weeks each of treatment will be administered, and a disease evaluation will be carried out after eight weeks, which completes one course of therapy. Surrogate endpoints to be measured include analysis of antigen-specific CTL response using a quantitative peptide-specific flow cytometry procedure (Dimer XI assay) and a quantitative intracellular staining assay. Both assays will be performed before, during, and after each treatment course. The portable pump will be worn on the belt and patients will be permitted to ambulate during the four-day infusion.

Eligible patients who are HLA-A2 positive will have a staging workup consisting of CT scans of chest, abdomen and pelvis and MRI scan of the head prior to initiation of therapy to define the extent of their disease.

No life threatening side effects or deaths were seen with previous tests of vaccines such as peptides, or dendritic cells pulsed with peptides or tumor lysates injected intralymphnodally in patients with metastatic melanoma. The toxicities related to injection of DNA vaccines subcutaneously or intravenously included headache, fevers, weakness, arthralgias and a rash that spontaneously resolved without therapy. In a recent and ongoing clinical trial of intranodal injection of a tyrosinase DNA plasmid, no severe or life-threatening side effects have been observed. In animal testing, the Synchrotope MA2M DNA plasmid vaccine had little significant toxicity. It is possible, but unlikely, that vaccination with the DNA plasmid vaccine in this trial may induce retinitis and even cause blindness due to inflammation in retinal pigment cells expressing Melan-A/MART-l. Melan-A/MART-l is also present on normal human

melanocytes, so it is possible that vaccination with the DNA plasmid vaccine may induce areas of vitiligo. Arthritis, pain, rashes, and kidney or liver dysfunction might also occur.

It is possible that there might be damage to the lymph node into which the DNA plasmid vaccine infusion is given. The lymph node might become edematous or tender, or bleeding may occur. This has been shown to be temporary, with the lymph node returning to normal after the injections. The plastic catheter will be inserted in a sterile manner into the lymph node, but infection at the injection site might also occur.

The staging tests, which showed evidence of disease, will be repeated at the end of a course of treatment at week eight. Patients with evidence of tumor regression may be re-treated with one subsequent course of therapy.